

Monosubstituted Reactive Derivatives of Metalloporphyrins for Phosphorescent Labeling of Biomolecules

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Abstract

In this poster we introduce new monofunctional and phosphorescent labeling reagents that are based on Platinum(II) and Palladium(II) Coproporphyrins and isothiocyanate reactive group. The labeling reagents are stable and pure chemical compounds that can be reacted smoothly with biomolecules that contain amino functional groups. The resulting bioconjugate can be used as a phosphorescent probe in bioanalytical systems in combination with time-resolved luminescence detection.

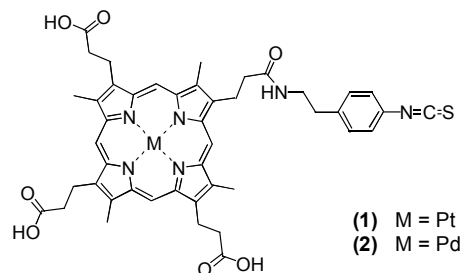


Fig 1. Chemical structure of the monofunctional phosphorescent labeling reagents with isothiocyanate reactive group.

Synthesis and Chemical Properties

The labeling reagents (1) and (2) (Fig.1) are prepared in five steps starting from a commercial compound, coproporphyrin tetraalkyl ester. The overall yields of the five step synthetic procedures for the Pt- and Pd- labels are 30 % and 50%, respectively.

The synthetic procedure affords the labeling reagents in pure form with exact chemical structure. The reagents are highly soluble in neutral or alkaline water and stable over years of storage. The reagents can be used for facile labeling of target molecules that contain one or more primary amino groups. The labeling reaction is carried out under slightly alkaline conditions (pH 9.5) and the reaction affords phosphorescent conjugates in high yields.

Photophysical Properties

The labels are excited either with violet or green range of visible spectrum and exhibit intense phosphorescence at the red range (Fig 2.). The photophysical parameters are summarised in Table 1.

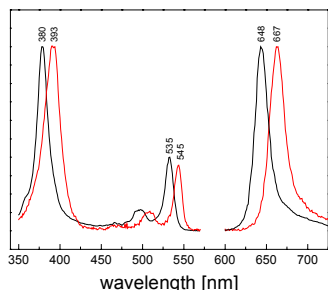


Fig.2. Excitation and emission spectra of the phosphorescent labeling reagents. Platinum -label (Black), Palladium -label (Red)

	Decay - Time	Quantum Yield	ϵ -value [M ⁻¹ cm ⁻¹]
Pt -label	100 μ s	~ 0.1	200 000
Pd -label	1000 μ s	~ 0.05	160 000

Table 1. Photophysical parameters of the Pt-and Pd-coproporphyrin labels.

Lowest limit of detection

By means of modulated green laser illumination (Nd-YAG, 532 nm, 10 mW cw) and a time-resolved luminescence detection, we have reached the following detection sensitivities:

- Pt-label 10^{-13} M
- Pd-label 10^{-12} M

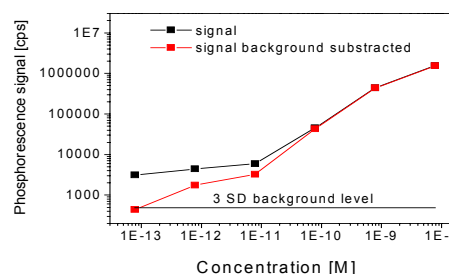


Fig. 3. Phosphorescence signal of the Platinum coproporphyrin label in deoxygenated aqueous solution of TX-100 (1%) measured with a time-resolved plate fluorometer (in house construction)

Bibliography:

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